

# BELOWGROUND PROCESSES IN NITROGEN FERTILIZED COTTONWOOD AND LOBLOLLY PINE PLANTATIONS

Kye-Han Lee and Shibu Jose<sup>1</sup>

**Abstract**—We measured soil respiration, fine root biomass production, and microbial biomass along a fertilization gradient (0, 56, 112, and 224 kg N ha<sup>-1</sup> per year) in 7-year-old cottonwood and loblolly pine plantations, established on a well-drained, Redbay sandy loam (a fine-loamy, siliceous, thermic Rhodic Paleudlt), in northwest Florida. Annual soil respiration rate was significantly greater in cottonwood (781 g C m<sup>-2</sup> per year) than in loblolly pine (692 g C m<sup>-2</sup> per year). Nitrogen (N) fertilization had a significant negative effect on soil respiration in cottonwood, but no effect was observed in loblolly pine stands. Annual fine root production was significantly greater in cottonwood (221 g m<sup>-2</sup> per year) than that in loblolly pine (144 g m<sup>-2</sup> per year) without N fertilization effect. Microbial biomass, however, was reduced by N fertilization in both species. These results suggest that belowground responses to fertilization can vary widely between conifers and hardwoods.

## INTRODUCTION

The potential response of aboveground biomass growth to fertilization is nearly always positive. However, belowground response to fertilization is often unclear in plantations and natural forests because of the uncertainty involved in quantifying root biomass. In addition to altering root growth and turnover rates, fertilization could also affect soil processes such as respiration, microbial activity, and soil pH. Nitrogen (N) fertilization of forest plantations has become increasingly important as an intensive management tool in recent years (Fox 2000). It has been speculated that intensive forest management may lead to a reduction in soil carbon (C), due partially to increased soil respiration (Harmon and others 1990). The few existing studies have found conflicting results, with one reporting an increase (Gallardo and Schlesinger 1994) and the other reporting a decrease (Haynes and Gower 1995). Since soil respiration results from two main sources, root respiration and the decomposition of organic matter and associated respiration of soil fauna, the conflicting results reported could be the result of fertilizer-induced differences in C fixation and allocation patterns among different tree species (Raich and Tufekcioglu 2000). Belowground C input through fine root production and turnover and associated microbial activity are well-documented processes in the C and nutrient cycling of forest ecosystems (Keyes and Grier 1981, McLaugherty and others 1982). Majdi and Kangas (1997) reported that a high input of N increased fine root mortality and decreased production and longevity, while addition of N-free fertilizer extended fine root longevity. In addition to the amount of belowground carbon, soil respiration could also be influenced by a number of other parameters, including the inherent and/or fertilizer-induced variation in soil physical and chemical properties, which may influence the quality and quantity of soil fauna. Microbial biomass has been shown to be a sensitive indicator responding quickly to environmental impacts. Zhang and Zak (1998) found that N fertilization increased microbial biomass and root growth. However, others have reported decreased soil microbial biomass in response to high rates of N fertilization (Smolander and others 1994). These conflicting results

may be due to the differences in the initial status of the microbial communities, soil pH, organic matter, and soil nutrient contents. Though fairly large amounts of information exist on how forest stands and individual trees respond to nutrient input, many uncertainties and controversies exist on the belowground responses and on the interactions among various processes such as soil respiration, fine root production and microbial biomass in response to long-term fertilization. We utilized a 7-year-old fertigation trial with cottonwood (*Populus deltoides* Marsh.) and loblolly pine (*Pinus taeda* L.) to investigate the following questions: (1) how do soil respiration, microbial biomass, fine root production, and soil pH vary for hardwood vs. conifer stands (established on the same soil) along a N fertilization gradient, and (2) what are the interrelationships among the above mentioned variables and how do they vary between species?

## MATERIALS AND METHODS

### Study Site

This study was conducted in 7-year-old cottonwood and loblolly pine plantations established in 1995 on an agricultural field (16.2 ha) located in northern Santa Rosa County, FL (30°50' N, 87°11' W), U.S.A. The climate is temperate with mild winters and hot, humid summers. The soil is characterized as a well-drained, Redbay sandy loam (a fine-loamy, siliceous, thermic Rhodic Paleudlt). A drip irrigation system was run to apply water and N fertilizer (0, 56, 112, 224 N kg ha<sup>-1</sup> per year) in the treatment plots during growing seasons for 8 consecutive years from 1995 to 2002. Three plots in each treatment were selected for this experiment.

### Soil Respiration

We measured soil respiration monthly using the soda-lime technique (Edwards 1982) from June 2001 through May 2002 at three randomly selected locations in each plot. The measurements were conducted at the same locations during the study year. Cylindrical plastic buckets, 20 cm tall and 27.5 cm in diameter, were used as measurement chambers. An uncovered tin cup containing 60 g of oven-dried

<sup>1</sup> Post-Doctoral Research Associate and Assistant Professor, School of Forest Resources and Conservation, University of Florida, 5988 Highway 90, Bldg. 4900, Milton, FL 32583, respectively.

soda-lime was placed under the inverted plastic chamber. After a 24-hour CO<sub>2</sub> absorption period, the tin cup was removed and oven dried at 105 °C for 24 hours to measure weight gain. Soil temperature and moisture were measured at 12 cm depth adjacent to each chamber during installation.

### Fine Root Production

Monthly fine root (< 2 mm) production was determined by means of ingrowth core method at three randomly selected locations in each plot from June 2001 through May 2002. The ingrowth core method consists of removing a core to a depth of 30 cm by a sharp-edge steel corer with an internal diameter of 5.1 cm and refilling the hole with root free soil, which was taken from the same site and sieved through a 2 mm screen. The core was resampled after a month and the roots that had grown into the core counted as fine root production. The same hole was used for ingrowth sampling during the entire study year. Soil cores were wet sieved through a fine mesh screen, and all hand-sorted root fragments were considered fine root production. Roots were dried at 65 °C for 48 hours and weighed to  $\pm 0.1$  mg.

### Microbial Biomass C

Soil microbial biomass C was measured by chloroform fumigation-extraction procedure (Vance and others 1987). Three soil cores (10 cm deep) were taken near the location for soil respiration measurement in each plot in September 2001. These soil samples were used for microbial biomass C, soil organic matter, and soil pH measurements. Sieved 50 g soil samples were placed in a 100 mL beaker and a 250 mL HDPE bottle for the fumigated and control samples, respectively. The fumigated samples were incubated in ethanol-free chloroform in evacuated desiccators for 24 hours at 25 °C. Fumigated and control samples were extracted with 100 mL of 0.5 M K<sub>2</sub>SO<sub>4</sub>, shaken for 1 hour, and filtered through Whatman No. 42 filter paper into 60 mL HDPE bottles. The extracted samples were acidified with phosphoric acid and frozen until analysis for dissolved organic C. Soil organic matter was determined by the ignition method using a 5 g dried soil, and soil pH was measured using a 1:5 soil-deionized water paste.

### Statistical Analysis

The effect of treatments (species and fertilization) on soil respiration rate and fine root production was tested using an unequal two-way ANOVA; microbial biomass C, soil organic matter, and pH was tested using the General Linear Model procedure of SAS (SAS Institute, Cary, NC). If significant treatment effects were revealed ( $\alpha = 0.05$ ), Tukey's studentized range test was used for mean separation. Possible effects of soil microbial biomass C, fine root production, soil organic matter, and soil pH on annual soil respiration rates were evaluated using Pearson correlation analysis (SAS Institute 1990).

## RESULTS AND DISCUSSION

### Soil Respiration

Annual soil respiration rates in this study ranged from 725 - 858 g C m<sup>-2</sup> per year (average of 781 g C m<sup>-2</sup> per year) under cottonwood to 647 - 720 g C m<sup>-2</sup> per year (average of 692 g C m<sup>-2</sup> per year) under loblolly pine stands and

differed significantly ( $P = 0.002$ ) between cottonwood and loblolly pine stands (table 1). N fertilization significantly affected soil respiration in cottonwood stands ( $P = 0.008$ ) but had no effect in loblolly pine stands (figs. 1A, 1B). Similar ranges have been reported in a number of studies. Raich and Schlesinger (1992) reported a mean soil respiration of 647 and 695 g C m<sup>-2</sup> per year from a wide array of temperate deciduous forests and coniferous forests, respectively. Annual soil respiration rate in loblolly pine was approximately 11 percent lower than that in cottonwood stands in our study, similar to a 10 percent reduction observed by Raich and Tufekcioglu (2000) while comparing deciduous and coniferous stands on identical soils. Temporal variation in soil respiration rates was closely related to soil temperature fluctuation in both species (fig. 2). Mean daily soil respiration rates displayed a significant exponential relationship with soil temperature; the relationship was stronger in cottonwood stands ( $R^2 = 0.81$ ) than in loblolly pine stands ( $R^2 = 0.51$ ) (fig. 3). Annual soil respiration rate in cottonwood stands was positively correlated with fine root biomass production ( $r = 0.64$ ) and soil microbial biomass C ( $r = 0.87$ ) and negatively correlated with soil pH ( $r = -0.81$ ). In loblolly pine stands, however, annual soil respiration was positively correlated with fine root biomass production ( $r = 0.54$ ) and soil organic matter content ( $r = 0.74$ ). Since root respiration and associated microbial activity are two major sources of soil respiration (Keyes and Grier 1981, McClaugherty and others 1982), it is not surprising that soil respiration decreased along an increasing N fertilization in cottonwood.

### Fine Root Production

Annual fine root production in cottonwood stands was 35 percent greater ( $P < 0.0001$ ) than that in loblolly pine stands (table 1). This agrees with results by Aber and others (1985) and Steele and others (1997), who showed that hardwood forests have greater fine root biomass production than pine forests of similar age. In a recent study, Usman and others (2000) reported that fine root production was 44.6 percent higher in oak forests than in nearby pine forests in central Himalaya. Despite having a decreasing trend of fine root production in cottonwood and increasing trend in loblolly pine along an increasing soil N fertilization, no significant differences among the fertilization treatments were observed in either species (fig. 1). Annual fine root production ranged from 208 to 241 g m<sup>-2</sup>

**Table 1—Means of soil respiration, fine root production, and microbial biomass in cottonwood and loblolly pine plantations in northwest Florida<sup>a</sup>**

Variable	Cottonwood	Loblolly pine
Soil respiration (g C m <sup>-2</sup> per year)	780.8 (19.3)a	691.5 (24.2)b
Fine root production (g m <sup>-2</sup> per year)	220.8 (28.5)a	144.2 (23.4)b
Microbial biomass (mg C kg <sup>-1</sup> dry soil)	144.3 (19.5)a	122.2 (25.7)a

<sup>a</sup> Numbers in parenthesis are the standard error of the mean (n = 12). Different letters within a row indicate significant differences (Tukey HSD test,  $\alpha = 0.05$ ).

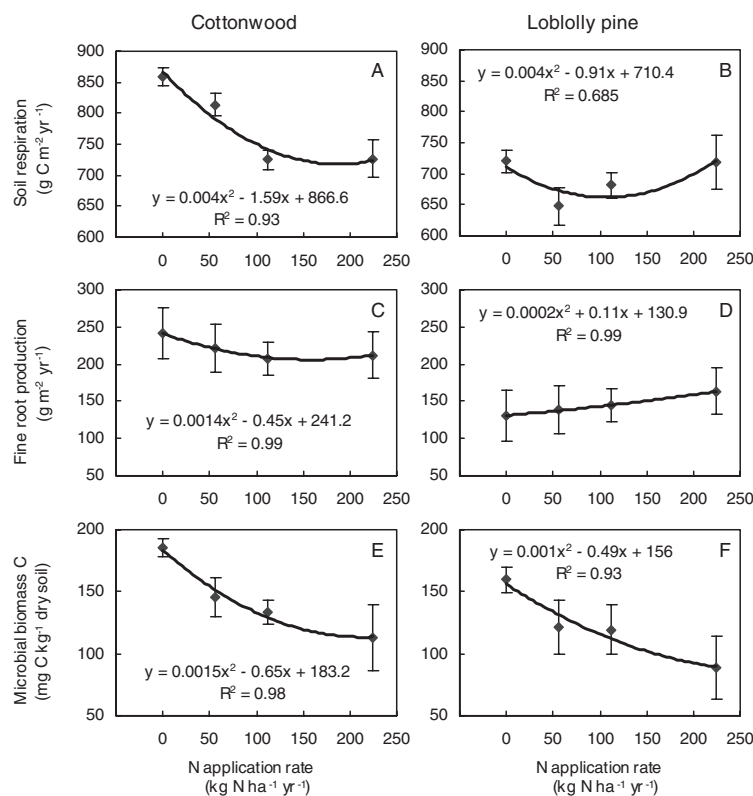


Figure 1—Effects of N fertilization rates on soil respiration (A and B), fine root production (C and D), and microbial biomass C (E and F) in cottonwood and loblolly pine plantations in northwest Florida. Vertical bars indicate standard error of the mean (n = 12).

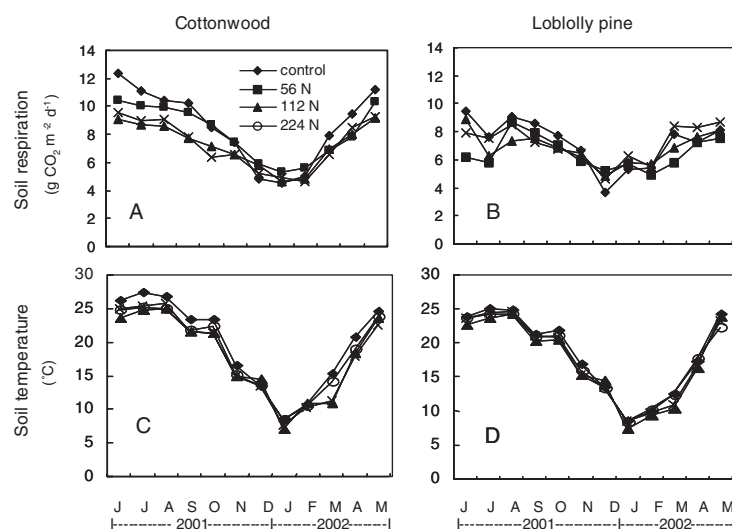


Figure 2—Mean monthly soil respiration rates in 7-year-old cottonwood (A) and loblolly pine (B) plantations along a fertilization gradient (0, 56, 112, and 224 kg N ha<sup>-1</sup> per year), and soil temperature (C and D) at 12 cm depth under both species.

per year in cottonwood stands, and from 131 to 163 g m<sup>-2</sup> per year in loblolly pine stands (figs. 1C, 1D). In comparison to other average fine root production estimates by ingrowth core methods, our estimation in cottonwood stands (221 g m<sup>-2</sup> per year) was within the range of

147-254 g m<sup>-2</sup> per year reported for temperate deciduous forests (Fahey and Hughes 1994, Hertel and Leuschner 2002); the 144.2 g m<sup>-2</sup> per year in loblolly pine stands was also within the range of 120-200 g m<sup>-2</sup> per year reported by Nadelhoffer and others (1985) and Aber and others (1985) for red pine plantations in Wisconsin.

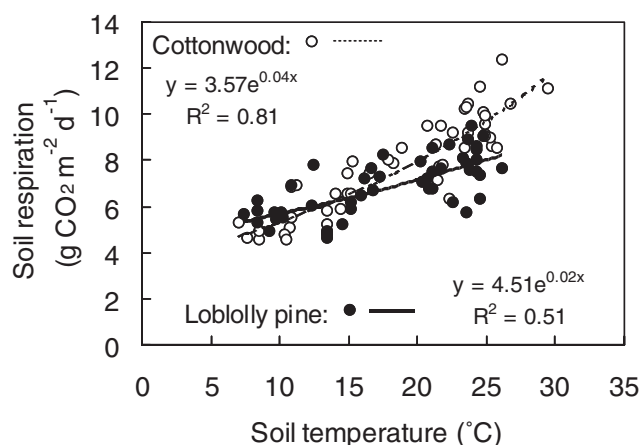


Figure 3—Relationship between soil respiration and soil temperature in 7-year-old cottonwood and loblolly pine plantations in northwest Florida.

### Microbial Biomass C

There was no difference in soil microbial biomass C between cottonwood and loblolly pine stands (table 1); however, N fertilization negatively affected soil microbial biomass C ( $P = 0.048$ ) in both species (figs. 1E, 1F). Similar results have been reported from other long-term fertilization trials (Fisk and Fahey 2001, Scott and others 1998). For example, Fisk and Fahey (2001) reported that microbial biomass C was significantly reduced by 8 years of continued N fertilization in a temperate hardwood forest. In contrast, shorter-term N fertilization has been shown to increase microbial biomass (Gallardo and Schlesinger 1994, Hart and Stark 1997). The immediate increase in microbial biomass following N additions suggests that N could be limiting in the soil. However, there is no obvious explanation for the decrease in microbial biomass over longer periods of treatment (Fisk and Fahey 2001). Although soil acidification and subsequent reduction in microbial biomass C could be suspected, we did not observe this for either species.

### CONCLUSIONS

Soil respiration decreased significantly along an increasing soil N gradient in cottonwood; however, it remained unchanged in loblolly pine. The speculation that fertilization might lower soil C due partially to increased soil respiration was found untenable in our experiment. Contrary to our expectation and some published reports, fine root production exhibited no significant differences among treatments in both species. There was a trend of decreasing fine root production in cottonwood and increasing production in loblolly pine along the increasing soil N gradient, which may become more pronounced in the future as stands get older. Averaged across all treatments, loblolly pine had 35 percent lower fine root production than cottonwood. Microbial biomass decreased along the increasing N gradient in both species and was correlated strongly to soil organic matter and pH. In general, our results suggest that long-term N fertilization can modify belowground soil processes in both hardwood and coniferous stands, but not always in identical ways.

### LITERATURE CITED

- Aber, J.D.; Melillo, J.M.; Nadelhoffer, K.J. [and others]. 1985. Fine root turnover in forest ecosystems in relation to quantity and form of nitrogen availability: a comparison of two methods. *Oecologia*. 66: 317-321.
- Edwards, N.T. 1982. The use of soda-lime for measuring respiration rates in terrestrial systems. *Pedobiologia*. 23: 321-330.
- Fahey, T.J.; Hughes, J.W. 1994. Fine root dynamics in a northern hardwood forest ecosystem, Hubbard Brook Experimental Forest, NH. *Journal of Ecology*. 82: 533-548.
- Fisk, M.C.; Fahey, T.J. 2001. Microbial biomass and nitrogen cycling responses to fertilization and litter removal in young northern hardwood forests. *Biogeochemistry*. 53: 201-223.
- Fox, T.R. 2000. Sustained productivity in intensively managed forest plantations. *Forest Ecology and Management*. 138: 187-202.
- Gallardo, A.; Schlesinger, W.H. 1994. Factors limiting microbial biomass in the mineral soil and forest floor of a warm-temperate forest. *Soil Biology and Biochemistry*. 26: 1409-1415.
- Harmon, M.E.; Ferrell, W.K.; Franklin, J.F. 1990. Effects of carbon storage of conversion of old-growth forests to young forests. *Science*. 247: 699-702.
- Hart, S.C.; Stark, J.M. 1997. Nitrogen limitation of the microbial biomass in an old-growth forest soil. *Ecoscience*. 4: 91-98.
- Haynes, B.E.; Gower, S.T. 1995. Belowground carbon allocation in unfertilized and fertilized red pine plantations in northern Wisconsin. *Tree Physiology*. 15: 317-325.
- Hertel, D.; Leuschner, C. 2002. A comparison of four different fine root production estimates with ecosystem carbon balance data in a *Fagus-Quercus* mixed forest. *Plant Soil*. 239: 237-251.
- Keyes, M.R.; Grier, C.C. 1981. Above- and belowground net production in 40-year-old Douglas-fir stands on low and high productivity sites. *Canadian Journal of Forest Resources*. 8: 265-279.
- Majdi, H.; Kangas, P. 1997. Demography of fine roots in response to nutrient applications in a Norway spruce stand in Southwestern Sweden. *Ecoscience*. 4: 199-205.
- McLaugherty, C.A.; Aber, J.D.; Melillo, J.M. 1982. The role of fine roots in the organic matter and nitrogen budgets of two forested ecosystems. *Ecology*. 63: 1481-1490.
- Nadelhoffer, K.J.; Aber, J.D.; Melillo, J.M. 1985. Fine roots, net primary production, and soil nitrogen availability: a new hypothesis. *Ecology*. 66: 1377-1390.
- Raich, J.W.; Schlesinger, W.H. 1992. The global carbon dioxide flux in soil respiration and its relationship to vegetation and climate. *Tellus*. 44B: 81-99.
- Raich, J.W.; Tufekcioglu, A. 2000. Vegetation and soil respiration: Correlations and controls. *Biogeochemistry*. 48: 71-90.
- Scott, N.A.; Parfitt, R.L.; Ross, D.J.; Salt, G.J. 1998. Carbon and nitrogen transformations in New Zealand plantation forest soils from sites with different N status. *Canadian Journal of Forest Research*. 28: 967-976.
- Smolander, A.; Kurka, A.; Kitunen, V.; Mälkönen, E. 1994. Microbial biomass C and N, and respiratory activity in soil of repeatedly limed and N- and P-fertilized Norway spruce stands. *Soil Biology and Biochemistry*. 26: 957-962.
- Steele, S.J.; Gower, S.T.; Vogel, J.G.; Norman, J.M. 1997. Root mass, net primary production and turnover in aspen, jack pine and black spruce forests in Saskatchewan and Manitoba, Canada. *Tree Physiology*. 17: 576-587.
- Usman, S.; Singh, S.P.; Rawat, Y.S.; Bargali, S.S. 2000. Fine root decomposition and nitrogen mineralisation patterns in *Quercus leucotrichophora* and *Pinus roxburghii* forests in central Himalaya. *Forest Ecology and Management*. 131: 191-199.
- Vance, E.D.; Brookes, P.C.; Jenkinson, D.S. 1987. An extraction method for measuring microbial biomass C. *Soil Biology and Biochemistry*. 19: 703-707.
- Zhang, Q.H.; Zak, J.C. 1998. Effects of water and nitrogen amendment on soil microbial biomass and fine root production in a semi-arid environment in West Texas. *Soil Biology and Biochemistry*. 30: 39-45.